The International Journal Of Engineering And Science (IJES)
|| Volume || 3 || Issue || 3 || Pages || 63-67 || 2014 ||
ISSN (e): 2319 – 1813 ISSN (p): 2319 – 1805

Sludge Characteristic and Pathogen Inactivation of Two Different Wastewater Treatment Plants in Antalya

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ABSTRACT

In this study, sludge from municipal and industrial waste water plants located in and around Antalya city, were sampled and analyzed for bacteriological, helminth and protozoon pathogens including Escherichia coli, Salmonella spp. helminth and protozoon species compositions were also determined. In addition the laboratory scale research was conducted to eliminate the microbiological pathogens. In this respect the effects of heat treatment and stabilization with lime were conducted. Results indicate that considerable amount of pathogens can be eliminated by heating the sludge up to 70°C whereas the addition of lime accelerates the termination the pathogens.

Keywords *- Liming and Heat treatment, Inactivation, Bacteria, Helminth, Sludge.*

Date of Submission: 11 March 2014 Date of Publication: 25 March 2014

I. INTRODUCTION

Waste sludge originating from wastewater treatment plants contain many human pathogens such as fecal coliform, fecal streptococci, Escherichia coli, Salmonella spp., protozoa cysts and helminth eggs, which threat to public health[1]. It is reported that the amount of sewage sludge produces annually in Europe is 6.5 million tons and 5.4 million tons in USA [2].

Both agriculture and industry can benefit from the use of sludge as either agricultural fertilizer or soil conditioner and offers a beneficial recycling route for biosolids [3]. Agricultural use is an interesting alternative if the health risks are taken in to account as the sludge may contain high concentrations of micro-organisms potentially pathogenic to humans and animals.[4]. Mawdsley et al. [5] reported that the survival of bacteria in the soil depends on many parameters, such as temperature, moisture, pH and other microorganisms. Also, Cools et al. [6] suggested that a low incubation temperature and high soil moisture aid survival of *E. coli* and *Enterococcus* sp. Therefore, it can be argued that the concentrations and the diversity of pathogens in waste water biosolids depend upon local climatic conditions in Mediterranean countries. Different techniques can be applied to reduce pathogen concentrations such as application of lime [CaO and $Ca(OH)_2$] [3] or pasteurization.

The goal of this research was to evaluate the physical, chemical and microbiological characteristics of sewage sludge sampled from two treatment plants in Antalya and to eliminate the pathogenic microorganisms with lime and heat treatment.

II. MATERIAL AND METHODS

2.1. Study site

In Antalya, two waste water treatment plants (WWTP) are in operation. The municipal waste water treatment plant of ASAT (500.000 equivalent population) is equipped with physical and biological treatment facilities. The second waste water treatment plant is located in Organized Industrial Zone (OIZ). It has a daily treatment capacity 20.000 m^3. In both plants the sludge from the secondary clarifiers are collected in a tank then transferred to the belt filter press for dewatering. The operators of the plants are faced with serious difficulties regarding the disposal of the sludge. The sludge is deposited in the landfill.

2.2. Collection of sludge sample

Sludge samples were collected monthly from April 2004 to March 2005 at ASAT WWTP (S1), and monthly from May 2004 to April 2005 at OIZ WWTP (S2). In total, 22 samples of untreated sludge were collected directly from the sludge storage tanks. Two of the samples were not taken because of the some break down about from ASAT WTP. The sludge samples were collected at the sludge storage tank in plastic bottles (5 l) and transported in cool boxes to the laboratory. The sludge samples were air-dried prior to chemical analysis.

2.3. Microbial and Chemical assay of field samples

Dry matter (DM) was analyzed using the procedures outlined in the Standard and Methods for the Examination of Water and Wastewater ((7). Organic carbon was determined by oxidation with potassium dichromate and titration of excess dichromate with ammonium ferrosulfate [8]. Available-Phosphor in the 0.5 NaHCO3 extract was determined by the antimony-potassium-tartarate method [8]. Fecal coliform and fecal streptococci in sludge samples were determined using the membran filtration method [7]. The sludge sample is diluted in order to reach the number of bacteria ca. 10^4 to 10^6. *Salmonella* spp. and *E. coli* in the sludge samples were determined using the most probable number method (MPN) [9-10]. To detect *Salmonella* spp, the samples pre-enriched buffered in peptone water, where after the samples was inoculated to Rappaport-Vassiliadis Broth (RVB) and incubated at 37 °C for 18-24h. RVB was inoculated on Xylose-Lysine Desoxycholate agar (XLD) and the agar plates were incubated for 18-24h, after which Salmonella colonies were confirmed using Urea Broth (UB) and triple sugar iron agar (TSI). Before enumeration for *E coli*, pre-enriched samples, inoculated to Lauryl Tryptose Broth (LTB) and incubated at 37°C for 18-24h and Brilliant Green Bile Broth (BGBB) were inoculated with positive reaction samples, after which *E. coli* verified with INDOL test. *Salmonella* spp. and *E. coli* density is reported as MPN g-1 dry weight.

Protozoa cysts and helminth eggs in waste sludge samples were determined using the floating Bailenger Method [11]. Approximately 20 ml aliquot of each sample was processed for protozoa cysts and helminth eggs.

2.4. Sludge treatment by slaked lime and heat

After determination of the dry solids content of the sludge cake, it was treated with slaked lime which ensured a pH >12 throughout the experiments (4 wt % by weight of slaked lime per weight of sludge dry solids). Treatment times varied between 6-72 hours, after which sample of the limed sludge was analyzed for *Salmonella* spp., *E. coli*, protozoan cysts and helminth eggs.

III. RESULTS

Concentrations of *Escherichia coli*, *Salmonella* spp, protozoon cysts and helminth eggs determined periodically every month for one year and the mean of values summarized Table 1. During the study time, maximum and minimum of values dry matter and pH was between the range 0.7-1,2 % and 5.2-7.8 for S1 sludge, whereas in S2 sludge was it was between range of 0.5-1.0 % and 6.7-8.3. Result show that all parameters were higher in OIZ more than in ASAT, except the dry matter content.

Table 1. Characteristics of the raw sludge in S1 and S2 sludge. (Indicator microorganism values were based on Log 10).

	pH	DM (%)	Org. C (mg g^{-1} DM)	P (mg g^{-1} DM)	LOG-EC (MPN.g^{-1} DM)	LOG-SALM (MPN.g^{-1} DM)	LOG-PROT *10^4.g^{-1} DM	LOG-HELM *10^4.g^{-1} DM
S1*	7,0±0,3	1,2±0,2	227,3±3,6	460,9±50,7	4,2±0,1	4,2±0,1	3,4±0,1	2,6±0,3
S2*	7,4±0,1	0,7±0,04	231,0±1,6	1011,1±248,7	4,4±0,02	4,4±0,02	3,8±0,1	3,5±0,1

*S1: Sludge 1, ASAT WWTP; S2: Sludge 2, OIZ WWTP.

While the concentrations of helminth were higher in S2 than in S1, the protozoon concentrations were lower than S1. Possibility the higher of protozoons in S2 may be due to meat and milk processes. The species distribution in sludge is a function of prevalence of infection in the community health. During the analyses of protozoon cysts *Entamoeba coli, Naegleria spp.* and helminth eggs *Ascaris lumbricoides* were dominated in S1 and S2 sludge. For the both treatment plant the ratio of protozoon cysts and helminth eggs are summarized in Table 2.

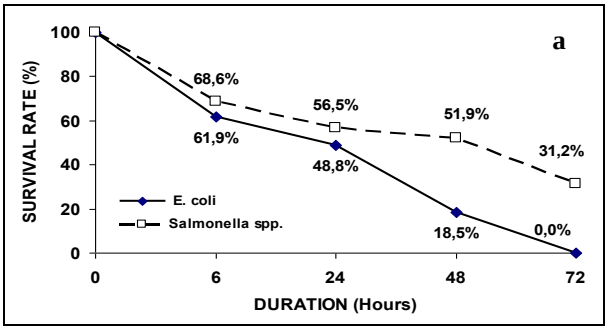
Table 2. Species composition and the ratio of Helminth eggs and Protozoa cysts (%) of helminth eggs and protozoon cysts in S1 and S2 sludge.

HELMINT			**PROTOZOON**		
Species	**S1(%)**	**S2(%)**	**Species**	**S1(%)**	**S2(%)**
Ascaris lumbricoides	37,3	30,7	*Entamoeba coli*	78,0	67,9
Hymenolepis nana	22,3		*Entamoeba hartmani*	2,6	4,3
Hymenolepis diminuta		8,3	*Naeglaria sp.*	19,4	27,2
Taenia saginata	10,8	-	*Giardia lamblia*	-	0,6
Taenia solium		8,9			
Opisthorchis fleneus	-	24,4			
Enterobius vermicularis	-	13,1			
Others*	29,6	14,6			
TOTAL group % in Sludge	23,7	35,9		76,3	64,1

*Others S1: 3.2% *Trichuris trichiura,* 3% *Taenia solium,* 8.9 %, *Capillaria philippinensis,* 3.8 %*Diphyllobothrium latum,* 8.0 % *Colonorchis sinensis,* 5.5 % *Enterobius vermicularis*)
*Others S2: 4.2% *Heterophes heteroppyes,* 1.6% *Schistosoma haematobium,* 1.5% *S. japonicum,* 7.4% *S.mansoi,* % 3

3.1 Sludge treatment with slaked lime

The inactivation curves of sludge for *Salmonella* spp., *E. coli,* are presented in Fig.1a. Comparison of these figures shows that pathogens are decreased to limit values after 72 hours by slaked liming. Contaminated slaked lime in sludge, 69% of *Salmonella* sp. still survived after 72 hours of treatment. However, to reach a negligible level of Ascaris eggs in contaminated sludge treated with slaked lime required a time more than 72 hours for sludge (Fig. 1b). Sample of the limed sludge and heat treatment was analyzed for *Salmonella* spp., *E. coli,* protozoan cysts and helminth eggs. Also, the inactivation rate of considered here, because the eggs of *Ascaris* were more resistant than those of *Trichuris* sp. and *Toxocara* sp.



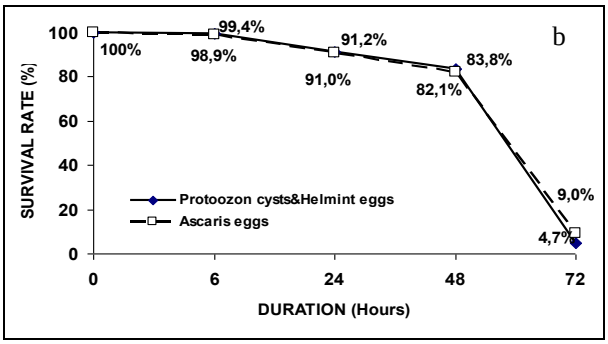


Figure 1. Survival of *Salmonella* sp. and *E. coli* (a) and Helminth eggs and Protozoon cyst (b) during the slaked liming

3.2 Sludge treatment with heat

For the heat treatment, sludge sample were heated between temperatures of 45^0C to70^0C and analyzed for each of the microbial parameter. Most of the temperatures are not effective for the treatment. Inactivation curves obtained for 65°C and 70°C showed in Fig. 2a, Fig 3b for *Salmonella spp*. and *E. coli.* The result indicates that the inactivation ratio was not affected to less than 55°C (Fig 3a and Fig 3b).

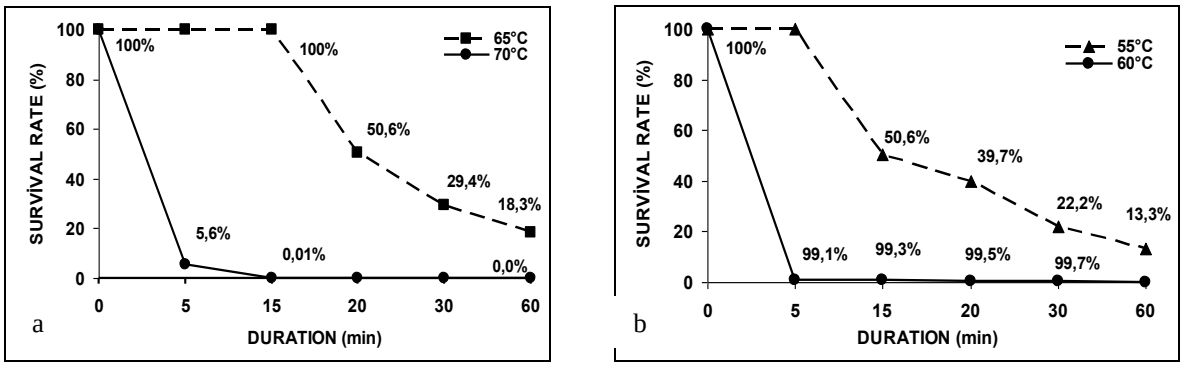


Figure 2. Survival of *Salmonella* spp. (a) and *E. coli* (b) with heat.

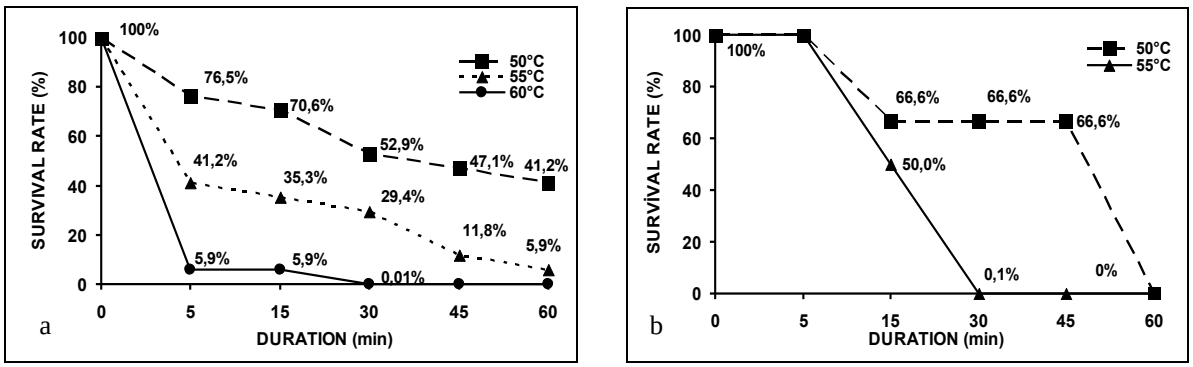


Figure 4. Treatment for Helminth eggs (a) and Protozoon cyst (b) with heat.

IV. DISCUSSIONS AND CONCLUSION

In this study, effects of heat and slaked lime on the pathogens of sludge under controlled laboratory conditions are demonstrated. Others studies focused on the pH effect of liming on pathogen inactivation, while only few investigated the inactivation bacteria, helminth eggs and protozoon cysts by combining the lime and temperature effect. The aim was to identify the treatments that produce sanitized sludge, with undetectable levels of parasites and bacteria, which can therefore be used in agriculture. Research was conducted at two wastewater treatment plants, ASAT WTP and OIZ WTP.

The result of bacteriological and parasitological analysis are consistent with those of Estrada et al. [12], Sahlström et al. [13], Gantzer et al. [4], Amahmaid et al. [14], Sidhu et al. [15], Carrington et al. [16], All the pathogen levels are less than our values. If sludge containing pathogens are used in tropical soil applications there is a danger of transmittance of disease and parasite infections. Ascaris eggs have been able to survive in soil for at least 7 years; Salmonella, up to 112 days and cysts of Entamoeba, for 8 days [17, 18]. In order that, the level of pathogens decreases, we treated the sludge with two methods. Heat treatment 49% of Salmonella removed after 30 minutes at 65°C and all of Salmonella removed after 15 minutes at 70°C. With slaked liming, 71% of Salmonella removed in 24 hours. At the end of the 72 hours 98% of Salmonella removed.

With the heat treatment, *E. coli* started to remove after 60 minute at 550C and all of them removed in one hour at 60°C. When E. coli treated with slaked lime 98% of *E. coli* removed in 6 hours and all of them are removed at the end of 72 hours. In recent years some researchers reported that helminth eggs [3, 17] and pathogen bacteria [4] all removed with slaked lime by heating at higher than 50°C in one hour. In our study we removed 50% of helminth eggs and protozoa cysts in 24 hours and all of them are removed at the end of the 72 hours. This study has demonstrated that with heat treatment all pathogens remove in one hour at 70°C and with slaked liming, if pH is higher than 12 all pathogens remove in 72 hours.

V. ACKNOWLEDGEMENTS

The authors are grateful to Akdeniz University for the financial support of this project (No:2004.02.0121.003).

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